

PCT

RAW SEQUENCE LISTING DATE: 07/16/2004
PATENT APPLICATION: US/10/501,071 TIME: 16:37:40

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

```
5 <110> APPLICANT: University of Newcastle Upon Tyne
     9 <120> TITLE OF INVENTION: Fusion Proteins
     13 <130> FILE REFERENCE: 43952/JMD/MAR
C--> 17 <140> CURRENT APPLICATION NUMBER: US/10/501,071
C--> 17 <141> CURRENT FILING DATE: 2004-07-09
     17 <150> PRIOR APPLICATION NUMBER: GB 0200689.8
     19 <151> PRIOR FILING DATE: 2002-01-10
     23 <160> NUMBER OF SEQ ID NOS: 61
     27 <170> SOFTWARE: PatentIn version 3.1
     31 <210> SEQ ID NO: 1
                                                               ENTERED
     33 <211> LENGTH: 9
     35 <212> TYPE: PRT
     37 <213> ORGANISM: Artificial Sequence
     41 <220> FEATURE:
     43 <223> OTHER INFORMATION: Ala3-His6 tail
     45 <400> SEQUENCE: 1
     47 Ala Ala Ala His His His His His
     51 <210> SEQ ID NO: 2
     53 <211> LENGTH: 25
     55 <212> TYPE: PRT
     57 <213> ORGANISM: Escherichia coli
     61 <400> SEQUENCE: 2
     63 Met Asn Met Lys Lys Leu Ala Thr Leu Val Ser Ala Val Ala Leu Ser
                      5
     67 Ala Thr Val Ser Ala Asn Ala Met Ala
                  20
     71 <210> SEQ ID NO: 3
     73 <211> LENGTH: 5
     75 <212> TYPE: PRT
     77 <213> ORGANISM: Artificial Sequence
     81 <220> FEATURE:
     83 <223> OTHER INFORMATION: Cleavage site for enterokinase
     85 <400> SEQUENCE: 3
     87 Asp Asp Asp Lys
     91 <210> SEQ ID NO: 4
     93 <211> LENGTH: 4
     95 <212> TYPE: PRT
     97 <213> ORGANISM: Artificial Sequence
     101 <220> FEATURE:
     103 <223> OTHER INFORMATION: Cleavage site for thrombin.
```

105 <400> SEQUENCE: 4

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

107 Leu Val Pro Arg 108 1 111 <210> SEQ ID NO: 5 113 <211> LENGTH: 4 115 <212> TYPE: PRT 117 <213> ORGANISM: Artificial Sequence 121 <220> FEATURE: 123 <223> OTHER INFORMATION: Cleavage site for factor Xa 125 <400> SEQUENCE: 5 127 Ile Glu Gly Arg 128 1 131 <210> SEQ ID NO: 6 133 <211> LENGTH: 4 135 <212> TYPE: PRT 137 <213> ORGANISM: Artificial Sequence 141 <220> FEATURE: 143 <223> OTHER INFORMATION: 4xHis tag 145 <400> SEQUENCE: 6 147 His His His His 148 1 151 <210> SEQ ID NO: 7 153 <211> LENGTH: 5 155 <212> TYPE: PRT 157 <213> ORGANISM: Artificial Sequence 161 <220> FEATURE: 163 <223> OTHER INFORMATION: 5xHis tag 165 <400> SEQUENCE: 7 167 His His His His 168 1 171 <210> SEQ ID NO: 8 173 <211> LENGTH: 6 175 <212> TYPE: PRT 177 <213> ORGANISM: Artificial Sequence 181 <220> FEATURE: 183 <223> OTHER INFORMATION: 6xHis tag 185 <400> SEQUENCE: 8 187 His His His His His 188 1 191 <210> SEQ ID NO: 9 193 <211> LENGTH: 7 195 <212> TYPE: PRT 197 <213> ORGANISM: Artificial Sequence 201 <220> FEATURE: 203 <223> OTHER INFORMATION: 7xHis tag 205 <400> SEQUENCE: 9 207 His His His His His His 208 1 211 <210> SEQ ID NO: 10 213 <211> LENGTH: 8

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

```
215 <212> TYPE: PRT
217 <213> ORGANISM: Artificial Sequence
221 <220> FEATURE:
223 <223 > OTHER INFORMATION: 8xHis tag
225 <400> SEQUENCE: 10
227 His His His His His His His
228 1
231 <210> SEQ ID NO: 11
233 <211> LENGTH: 9
235 <212> TYPE: PRT
237 <213> ORGANISM: Artificial Sequence
241 <220> FEATURE:
243 <223> OTHER INFORMATION: 9xHis tag
245 <400> SEQUENCE: 11
247 His His His His His His His His
248 1
251 <210> SEQ ID NO: 12
253 <211> LENGTH: 10
255 <212> TYPE: PRT
257 <213> ORGANISM: Artificial Sequence
261 <220> FEATURE:
263 <223> OTHER INFORMATION: 10xHis tag
265 <400> SEOUENCE: 12
267 His His His His His His His His His
268 1
                    5
                                        10
271 <210> SEQ ID NO: 13
273 <211> LENGTH: 93
275 <212> TYPE: PRT
277 <213> ORGANISM: Escherichia coli
281 <400> SEQUENCE: 13
283 Asn Asn Gly Ala Ser Gly Ala Asp Ile Asn Asn Tyr Ala Gly Gln Ile
287 Lys Ser Ala Ile Glu Ser Lys Phe Tyr Asp Ala Ser Ser Tyr Ala Gly
291 Lys Thr Cys Thr Leu Arg Ile Lys Leu Ala Pro Asp Gly Met Leu Leu
          35
                                40
295 Asp Ile Lys Pro Glu Gly Gly Asp Pro Ala Leu Cys Gln Ala Ala Leu
299 Ala Ala Ala Lys Leu Ala Lys Ile Pro Lys Pro Pro Ser Gln Ala Val
300 65
303 Tyr Glu Val Phe Lys Asn Ala Pro Leu Asp Phe Lys Pro
304
                    85
307 <210> SEQ ID NO: 14
309 <211> LENGTH: 348
311 <212> TYPE: PRT
313 <213> ORGANISM: Artificial Sequence
317 <220> FEATURE:
319 <223> OTHER INFORMATION: TolA-BCL fusion protein
321 <400> SEQUENCE: 14
```

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

323	Met	His	His	His	His	His	His	Ser	Ser	Asn	Asn	Gly	Ala	Ser	Gly	Ala
324					5					10		_	_		15	
	Asp	Ile	Asn	Asn	Tyr	Ala	Gly	Gln		Lys	Ser	Ala	Ile		Ser	Lys
328	1		_	20		•			25	-	m1	G	m1	30		-1.
	Pne	Tyr	_	Ala	ser	ser	Tyr		GIA	ьys	Tnr	Cys		ьeu	Arg	11e
332	T 1/0	T 011	35 71 2	Dro	7 cm	C111	Mot	40	LOU	Λαn	Tlo	Larc	45 Pro	Glu.	Clv	Clv
336	пур	50	Ala	Pro	Asp	GIY	55	пеп	пеп	Asp	116	Б Б Б Б	FIO	Giu	Gry	GIY
	Asp		Ala	Leu	Cvs	Gln		Ala	Leu	Ala	Ala		Lvs	Leu	Ala	Lvs
340					0,10	70					75		-1-			80
		Pro	Lys	Pro	Pro	Ser	Gln	Ala	Val	Tyr	Glu	Val	Phe	Lys	Asn	Ala
344			-		85					90				_	95	
347	Pro	Leu	Asp	Phe	Lys	Pro	Gly	Gly	Gly	Ser	Gly	Ser	Leu	Val	Pro	Arg
348				100					105					110		
	Gly	Ser	_	Pro	Ser	Gln	Ser		Arg	Glu	Leu	Val		Asp	Phe	Leu
352	_		115	_	_		_	120	_	_	_	_	125	_,	_	_
	Ser	_	Lys	Leu	Ser	GIn	_	GIY	Tyr	Ser	Trp		GIn	Phe	ser	Asp
356	1707	130	~1	Asn	7 ~~~	mb ~	135	. ה ר	Dwo	C1	C1	140	C1.,	Co~	C1,,	Mot
	145	Gru	GIU	ASII	Arg	150	GIU	АІА	PIO	Giu	155	1111	GIU	SET	Giu	160
		Thr	Pro	Ser	Ala		Asn	Glv	Asn	Pro		Trp	His	Leu	Ala	
364	0			501	165			0-1		170	-				175	
	Ser	Pro	Ala	Val		Gly	Ala	Thr	Ala	His	Ser	Ser	Ser	Leu	Asp	Ala
368				180		_			185					190	_	
371	Arg	Glu	Val	Ile	Pro	Met	Ala	Ala	Val	Lys	Gln	Ala	Leu	Arg	Glu	Ala
372	,	•	195					200					205			
	Gly	_	Glu	Phe	Glu	Leu	_	Tyr	Arg	Arg	Ala		Ser	Asp	Leu	Thr
376	_	210	_			— 1	215	~7	— 1			220		5 1	~ 1	01
		GIn	Leu	His	шe		Pro	GLY	Thr	Ala	_	GIn	ser	Pne	GIU	G1n 240
	225	V-1	λan	Glu	T 011	230 Pho	7.22	7 cn	Glw	17 a 1	235	Trn	G1v	λνα	т1Д	
384	vai	vaı	ASII	Giu	245	FIIC	Arg	Asp	GLY	250	ASII	пр	Gry	Arg	255	vai
	Ala	Phe	Phe	Ser		Glv	Glv	Ala	Leu		Val	Glu	Ser	Val		Lvs
388				260		2	1		265	-1-				270		-2 -
391	Glu	Met	Gln	Val	Leu	Val	Ser	Arg	Ile	Ala	Ala	Trp	Met	Ala	Thr	Tyr
392			275					280				-	285			
395	Leu	Asn	Asp	His	Leu	Glu	Pro	Trp	Ile	Gln	Glu	Asn	Gly	Gly	Trp	Asp
396		290		_			295			_	_	300	_			
		Phe	Val	Glu	Leu	-	Gly	Asn	Asn	Ala		Ala	Glu	Ser	Arg	
	305	~3	~7		51	310	•		D1	.	315	~1	14 - A	m1	**- 7	320
	_	Gin	GIU	Arg		Asn	Arg	Trp	Pne		Thr	GIY	Met	Thr		Ala
404		1727	Wal.	Leu	325	Gl ₁₇	Sor	Tau	Dho	330	71 200	Larc			335	
408	_	vai	vaı	340	пеп	GIY	Ser	пеп	345	Ser	AIG	цуъ				
		0> S1	EO II	ON C	: 15				- 13							
				H: 2:												
	<21															
				ISM:	Art	ific	ial s	Seque	ence							
421	<22	0 > F	EATU	RE:												

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

```
423 <223> OTHER INFORMATION: TolA-BCL fusion protein after thrombin cleavage
425 <400> SEQUENCE: 15
427 Gly Ser Arg Pro Ser Gln Ser Asn Arg Glu Leu Val Val Asp Phe Leu
431 Ser Tyr Lys Leu Ser Gln Lys Gly Tyr Ser Trp Ser Gln Phe Ser Asp
               20
                                    25
435 Val Glu Glu Asn Arg Thr Glu Ala Pro Glu Gly Thr Glu Ser Glu Met
439 Glu Thr Pro Ser Ala Ile Asn Gly Asn Pro Ser Trp His Leu Ala Asp
443 Ser Pro Ala Val Asn Gly Ala Thr Ala His Ser Ser Ser Leu Asp Ala
                                            75
447 Arq Glu Val Ile Pro Met Ala Ala Val Lys Gln Ala Leu Arq Glu Ala
451 Gly Asp Glu Phe Glu Leu Arg Tyr Arg Arg Ala Phe Ser Asp Leu Thr
               100
                                    105
455 Ser Gln Leu His Ile Thr Pro Gly Thr Ala Tyr Gln Ser Phe Glu Gln
           115
                                120
459 Val Val Asn Glu Leu Phe Arg Asp Gly Val Asn Trp Gly Arg Ile Val
                            135
       130
                                                140
463 Ala Phe Phe Ser Phe Gly Gly Ala Leu Cys Val Glu Ser Val Asp Lys
467 Glu Met Gln Val Leu Val Ser Arg Ile Ala Ala Trp Met Ala Thr Tyr
                    165
                                        170
471 Leu Asn Asp His Leu Glu Pro Trp Ile Gln Glu Asn Gly Gly Trp Asp
472
                180
                                    185
475 Thr Phe Val Glu Leu Tyr Gly Asn Asn Ala Ala Glu Ser Arg Lys
                                200
           195
479 Gly Gln Glu Arg Phe Asn Arg Trp Phe Leu Thr Gly Met Thr Val Ala
                           215
483 Gly Val Val Leu Leu Gly Ser Leu Phe Ser Arg Lys
484 225
                        230
487 <210> SEQ ID NO: 16
489 <211> LENGTH: 115
491 <212> TYPE: PRT
493 <213> ORGANISM: Artificial Sequence
497 <220> FEATURE:
499 <223> OTHER INFORMATION: Tagged TolAIII region of pTol vectors
501 <220> FEATURE:
503 <221> NAME/KEY: MISC FEATURE
505 <222> LOCATION: (107)..(111)
507 <223> OTHER INFORMATION: Xaa residues represent cleavage sites DDDDK (SEQ ID NO: 3),
508
          (SEQ ID NO: 4; no Xaa at position 111) or IEGR (SEQ ID NO: 5; no
         Xaa at position 111)
513 <400> SEQUENCE: 16
515 Met His His His His His Ser Ser Asn Asn Gly Ala Ser Gly Ala
519 Asp Ile Asn Asn Tyr Ala Gly Gln Ile Lys Ser Ala Ile Glu Ser Lys
                20
                                    25
```

LVPR

RAW SEQUENCE LISTING ERROR SUMMARY PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

Please Note:

Use of n and/or Xaa have been detected in the Sequence Listing. Please review the Sequence Listing to ensure that a corresponding explanation is presented in the <220> to <223> fields of each sequence which presents at least one n or Xaa.

Seq#:16; Xaa Pos. 107,108,109,110,111

Seq#:22; Xaa Pos. 14,15
Seq#:23; Xaa Pos. 13,14
Seq#:24; Xaa Pos. 13,14

VERIFICATION SUMMARY

PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt Output Set: N:\CRF4\07162004\J501071.raw

L:17 M:270 C: Current Application Number differs, Replaced Current Application No

L:17 M:271 C: Current Filing Date differs, Replaced Current Filing Date L:539 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:16 after pos.:96

L:667 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:22 after pos.:0 L:701 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:23 after pos.:0 L:731 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:24 after pos.:0

STATISTICS SUMMARY

PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

Application Serial Number: US/10/501,071

Alpha or Numeric or Xml: Numeric

Application Class:

Application File Date: 07-09-2004

Art Unit: PCT

Software Application: PatentIN3.1 Total Number of Sequences: 61

Total Nucleotides: 1032 Total Amino Acids: 2629 Number of Errors: 0 Number of Warnings: 4 Number of Corrections: 2

MESSAGE SUMMARY

270 C: 1 (Current Application Number differs)

271 C: 1 (Current Filing Date differs)

341 W: 4 ((46) "n" or "Xaa" used)